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The Effect of EM1 and Organic Fertilizers on Productivity, Protein, and Oil Content of Two *Zea mays. L* Varieties and the Importance of Environmental Sustainability

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Abstract

Sustainable agriculture requires increasing farmers' knowledge of methods and fertilizers beneficial to soil and plant health. This study explored alternatives to chemical fertilizers to reduce their overuse and restore soil health. This study evaluated the effects of organic fertilizer and EM1 biofertilizer on the growth, yield, and chemical composition of two *Zea mays* Linnaeus, 1753 varieties, hybrid and local, under field conditions. Four fertilizer treatments were applied: organic fertilizer, EM1, organic + EM1, and a control. The results showed significant differences among treatments for most agronomic and biochemical traits. The combined organic + EM1 treatment produced the highest values for leaf number, ear diameter, and grains per row in both varieties. The hybrid variety exhibited superior vegetative growth, while the local variety responded better in grain protein and oil content. The highest protein content was observed in the local variety under the organic + EM1 treatment (9.08 g), followed by EM1 alone (9.02 g), while the control had the lowest value (8.38 g). Similarly, oil content was highest under the combined treatment, reaching 8–9%, compared with 5–6% in the control. These improvements are attributed to the positive effects of organic matter and EM1 biofertilizers in enhancing soil fertility, nutrient availability, and microbial activity. Overall, the findings indicate that using organic and EM1 fertilizers significantly enhances maize growth, productivity, and grain quality while promoting soil fertility and environmental sustainability. Therefore, reducing reliance on chemical fertilizers and replacing them with organic and bio-based alternatives represents a viable strategy for sustainable maize production and long-term ecological balance.

Keywords

Productivity; Protein; Oil; Environmental sustainability

Introduction

Zea mays L*,* a cereal species in the Poaceae family, is an important crop due to its high productivity, adaptability to diverse environmental conditions, and ability to be grown twice a year (Al-Rumi, 2017). In terms of world production, maize ranks third after wheat and rice. However, maize is a nutrient-demanding crop, absorbing large amounts of nitrogen and other essential elements during growth (Ibrahim, 2013). This makes fertilization a critical component of maize production systems. Despite their effectiveness, long-term dependence on inorganic fertilizers has raised concerns due to their adverse impacts on soil health, including soil acidification, nutrient imbalance, reduced microbial diversity, and environmental pollution. In response, sustainable agricultural systems are increasingly shifting toward environmentally friendly fertilization strategies. Organic farming, supported by the use of organic amendments, enhances soil structure, improves nutrient availability, and reduces chemical residues in food (Al-Kalabi, 2018).

Using organic fertilizers (humus) instead of mineral fertilizers is one approach to reduce pollution caused by the use of synthetic mineral fertilizers. Humus is a rich source of nitrogen and phosphorus and contains humic and fulvic acids (Verkaik, 2006). It is a sustainable option to avoid the negative effects of chemical fertilizers on soil fertility in the long term (Avery, 2022). The addition of organic fertilizers has been shown to affect plant height, ear weight, and length (Kalay *et al*., 2020). The addition of organic matter has been shown to increase plant dry matter accumulation in maize and its uptake of macro- and micronutrients (Abdas *et al*, 2009). The use of organic fertilizers has enhanced maize growth and productivity, resulting in significant increases in total biomass and yield (Getachew *et al*, 2016). This indicates that organic options are essential for sustainable agriculture (Chen *et al*., 2025).

In sustainable agricultural production, the application of organic fertilizers is gaining increasing attention. Effective Microorganisms (EM1) biofertilizer is a natural biotechnological preparation consisting of compatible beneficial microbial consortia, including photosynthetic bacteria, lactic acid bacteria, actinomycetes, fungi, and yeasts (Nassour and Kbybo, 2022). These preparations are increasingly used in environmental protection, soil regeneration, and crop production (Cvijanović *et al*., 2024). Biofertilizers are environmentally friendly natural materials containing live microorganisms that are applied to seeds, soil, or plants to enhance nutrient availability and uptake (Hari, Seshadri and Perumal, 2010). Several researchers have demonstrated that EM1 application improves soil regeneration, nutrient cycling, crop growth, yield, and nutrient use efficiency (Cvijanović *et al*., 2024; Gewaily *et al*., 2016; Javaid and Mahmood, 2010; Karthik and Maheswari, 2021;).

Maize grains are rich in carbohydrates, proteins, oils, amino acids, and other biomolecules; therefore, improving grain yield and nutritional quality is essential to meet the growing global demand for food (Gao, 2020). Integrating organic fertilizers with EM1 biofertilizers may provide a synergistic approach to enhancing maize productivity while maintaining long-term soil fertility. The sustainable management of soil and agricultural resources is fundamental to achieving food security, land restoration, and environmental sustainability, as emphasized by the United Nations Sustainable

Development Goals aim to achieve (Atapattu *et al*, 2024). Therefore, evaluating the combined effects of organic and EM1 fertilizers on maize productivity is crucial for developing ecologically sustainable, low-input agricultural systems.

The importance of integrating organic fertilizers and EM1 biofertilizer lies in their ability to improve yield characteristics and crop quality, enhance soil fertility and nutrient utilization efficiency, thus contributing to reducing environmental impact and supporting sustainable agriculture. The importance of this study is also highlighted by the scarcity of local studies that have addressed the use of EM1 biofertilizer in conjunction with organic fertilizer in the studied area, which necessitates evaluating its impact on maize productivity and crop quality under local environmental conditions. Accordingly, the study aimed to:

1. Investigate the effect of organic fertilization and EM1 on improving grain quality and nutritional composition.
2. Evaluate the effect of using the biofertilizer EM1 alone and in combination with organic fertilizers on the growth and yield of maize.
3. Define the role of EM1 in improving nutrient use efficiency within sustainable farming systems.
4. Contribute to the development of environmentally friendly fertilization systems that support sustainable, low-input agricultural production.

Material and Methods

Experimental Design and Site

The field experiment was conducted using a Randomized Complete Block Design (RCBD) with three replicates for each treatment using organic fertilizer and EM1 fertilizer. Two seeds were planted in each hole. The planting density corresponds to an approximate area of 20 cm^2 per plant. Planting began on 7/8/2023 and will continue until 15/11/2023 (harvest stage), and a drip irrigation network (25 meters) was extended along each agricultural block with a drainage rate of 8 liters/hour.

Plant Materials: Plant Materials

Two varieties of *Zea mays* L. with different agronomic characteristics were used in this study. *Ghouta 82* (Local) were obtained from Crop Department at the General Commission for Scientific Agricultural Research (Damascus, Syria) is a locally adapted variety commonly cultivated in the study region for grain production. In contrast, *Jeta Poly* is a hybrid variety grown in limited areas mainly for forage purposes due to its shorter growth cycle. Ghouta 82 has a growth duration of 115–125 days from planting and an average yield of 5,720 tons/ hectare (Alsaeedi, Albalkhi and Alzoubi, 2022). *Jeta Poly* has a productive life of 90–100 days, yellow kernels, one to two ears per plant, and an average yield of 800 kg per 1000 m^2 (Golden Seeds, n.d.).

Fertilization Treatments

Compost was thoroughly mixed with the soil in the experimental plots before planting. Three treatments were applied (i) O_0 (Control): No organic fertilizer added, (ii) O_1

(Organic fertilizer): Applied at a rate of 20 mgha^{-1} (Al-Kalabi, 2018), and (iii) O_2 (EM1 fertilizer). The EM1 bio-enricher, containing a consortium of photosynthetic bacteria, yeasts, lactic acid bacteria, and fungi, was added to the soil through the irrigation water. The biofertilizer was applied three times, beginning from the activation stage, with one-month intervals between applications, at a rate of 750 cm^3 (per 1000 m^2).

Planting and Growth Conditions

On August 7, 2023, yellow maize (*Zea mays* L.) seeds were planted at a rate of two seeds per hole, with an average spacing of 20 cm between holes. By September 2, 2023, seedlings had reached an approximate height of 15 cm. The emergence of female inflorescences began on November 20, 2023, approximately 5–6 days after the appearance of male inflorescences.

Agronomic Measurements

The following growth and yield parameters were recorded:

(i) Average ear diameter (cm): Measured at both the largest and smallest parts of the ear using a measuring tape.
(ii) Number of grains per row per ear: Calculated by counting the grains in a single row of three randomly selected ears from each plot, and the arithmetic mean was recorded (Al-Shammari, Balkhi and Al-Zoubi, 2022).
(iii) Number of leaves per plant (leaf plant^{-1}): The total number of leaves was counted from the appearance of the first green leaf above the soil surface up to the top of the plant.
(iv) Number of ears per plant: The total number of ears formed on each plant was recorded.

Chemical Analysis of Kernels

Protein Extraction and Quantification: The protein content of corn kernels (cob) was determined following sequential solvent extraction in figure 1. Approximately 0.3 g of dried kernels were ground and extracted with boiling ethanol, and centrifuge for 5 minutes. Sequentially wash the residue with 96% ethanol, 85% acetone, 1:1 ethanol–acetone mixture, and 5% trichloroacetic acid (TCA; allow 20 min refrigeration before centrifugation). After each wash, centrifuge for 5 min/5000 rpm and discard the supernatant. Dry the final residue in an electric dryer. Add 10% KOH, mix, and incubate at 37°C for 1 hour. Filter and adjust the pH using HCl to prevent interference with the Coomassie reagent. Protein concentration was determined using the Coomassie dye-binding assay.

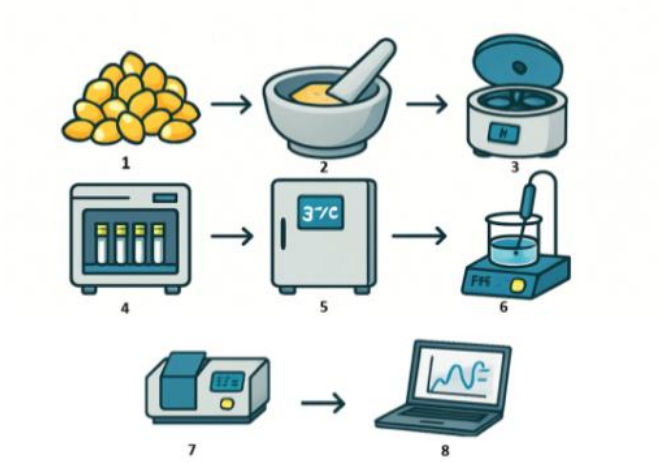

Figure 1: Illustrative figure showing Casein series for measuring protein content in corn kernels: (1) Preparing corn grain samples, (2) Grinding, (3) Oven (drying), (4) Cooling samples to 37°C, (5) pH adjustment, (6) Spectrophotometry, and (7) analyzing results.

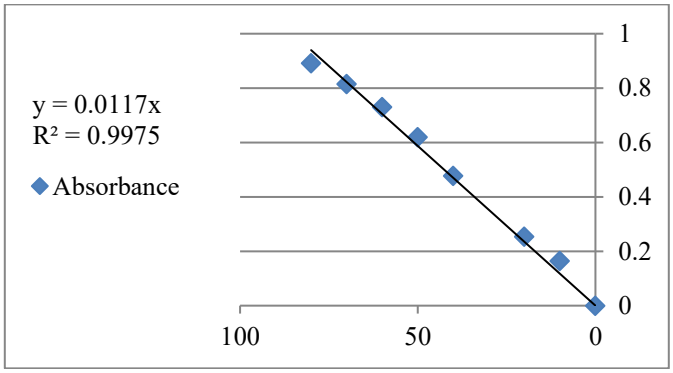

Figure 2: Casein calibration series used for protein quantification in maize kernels

Determination of Grain Oil Content

Oil Content of Corn Kernels (cob): Corn kernels were taken from each treatment to determine oil content using the Soxhlet method, 20 g per sample of fertilizer treatments, for 6 hours, hexane solvent method, according to (Ramadan *et al.*, 2025).

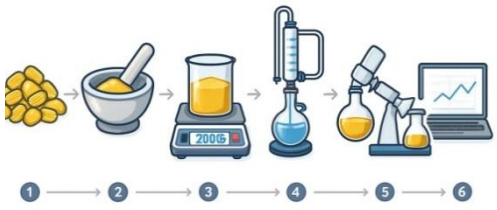

Figure 3: Illustrative figure showing Oil extraction: (1) Sample collection, (2) Grinding, (3) Weighing, (4) Oil extraction (Soxhlet apparatus), (5) Solvent evaporation (Rotary evaporator), (6) Oil quantification.

Statistical Analysis

All experiments were conducted using a randomized design with three replications per treatment. Data were statistically analyzed using Genstat software. A two-way analysis of variance (ANOVA) was employed to evaluate the effects of fertilizer treatments, maize varieties, and their interactions on the measured parameters. Means were compared to determine statistically significant differences, using a significance threshold of $P < 0.05$, corresponding to a 95% confidence level.

Results and Discussion

Effect of Fertilizer Treatments on Vegetative Growth

Number of leaves per Plant (leaf/plant): The results presented in Table 1 and Figure 4 indicate that there were significant differences in the mean number of leaves per plant for the two maize varieties, hybrid and local, under four fertilizer treatments: organic, organic+ EM1, EM1, and control. For the hybrid variety, the EM1-organic fertilizer treatment recorded the highest mean number of leaves (14 leaves/plant), indicating its high efficiency in promoting vegetative growth. This was followed by the organic treatment (14 leaves/plant), EM1 treatment (13.33 leaves/plant), and the control (11.67 leaves/plant). These results clearly indicate that the hybrid variety consistently produced a higher number of leaves when fertilized compared with the control. The local variety exhibited a similar pattern but with slightly higher values under the combined treatment. The organic + EM1 treatment resulted in the highest mean leaf number (15.33 leaves/plant). The organic and EM1 treatments each produced 13 leaves/plant, showing no significant difference between them. As expected, the control treatment had the lowest leaf number (11.67 leaves/plant). Overall, the combined organic + EM1 treatment consistently enhanced leaf production in both varieties. This improvement is likely due to synergistic effects between organic matter, which improves soil structure and nutrient availability, and EM1 microorganisms, which stimulate soil biological activity, thereby supporting vegetative growth.

Table 1: Number of leaves per plant in hybrid and local maize under different fertilizer treatments

Items	*Fertilizer Transactions*				*Mean*
	O_1	O_1*O_2	O_2	O_0	
Hybrid	14	16	13.33	11.67	13.75
	±1.26	±1.25	±1.24	±1.70	
Local	13	15.33	13	11.67	13.25
	±1.41	±0.47	±1.41	±1.24	
Mean	13.5	15.67	13.17	11.67	
L.S.D. $_1$	NS				
L.S.D.$_2$	1.803				
L.S.D.$_3$	NS				

O_1 = Organic fertilizer, O_2 =EM1, O_3 = Control, L.S.D. = Least Significant Difference at probability level0.05, L.S.D.$_1$ = Interaction, L.S.D.$_2$ = Fertilizer treatments, L.S.D.$_3$ = Cultivar, NS = Non-significant

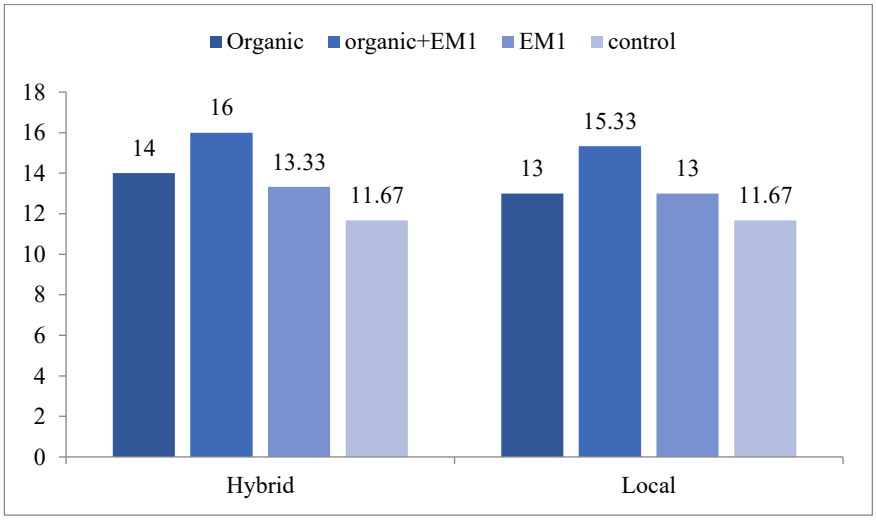

Figure 4: Average number of leaves per plant (leaf/plant) of hybrid and local maize as affected by different fertilizer treatments.

The number of leaves usually corresponds to the number of nodes on the stem, since each node bears a leaf (Al-Sahouki, 1990). The number of leaves produced by plants is also closely related to the factors that determine the initiation of ear formation, which shows a high degree of genetic variability (Hassanein, 1995). The application of organic fertilizer increased the number of leaves due to the role of humic acids contained in the organic material, which stimulates enhances plant vegetative growth (Thilakarathna and Upadhyay, 2024).This may also be attributed to the role of organic fertilizers in improving soil fertility, enhancing microbial activity, and increasing the availability of nutrients released during decomposition. Such improvement in root growth positively affects the increase in the number of leaves per plant (Abudurezike *et al*., 2025). These results could also be explained by the essential nutrients present in organic fertilizers, which enhance photosynthetic efficiency and improve the meristematic and physiological activities of the plants (Abd El-Gawad and Morsy, 2017). The EM1 fertilizer also played a significant role, as the EM1 treatment produced the highest number of leaves (Llamelo *et al*., 2016).

Similarly, the strong performance of EM1 treatments can be explained by the beneficial microorganisms that enhance nutrient cycling, stimulate plant growth regulators, and promote bud activity responsible for producing leaves and lateral branches (El-Shabasi, Gaafer and Zahran, 2003). Comparable increases in leaf number associated with organic and EM1 fertilization have been documented in previous studies (Faqira and Al-Shaibi, 2015), supporting the consistency of the present findings.

Effect of Fertilizer Treatments on Reproductive Traits

Number of ears/plant: The results presented in Table 2 indicate that there were no significant differences in the mean number of ears per plant for the two maize cultivars, hybrid and local, under four fertilizer treatments: organic, bio-organic, EM1, and control. For the hybrid cultivar, the bio-organic and organic fertilizer treatments recorded the highest mean number of ears per plant (1.66 ears/plant), followed by the EM1 treatment

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(1.33 ears/plant), while the control treatment recorded the lowest value (1 ear/plant), as shown in figures 5 and 6.

Similarly, the local cultivar exhibited a comparable pattern, where the organic, EM1, and bio-organic treatments all resulted in one ear per plant on average, whereas the organic treatment alone reached 1.33 ears/plant. These results suggest that ear formation in the studied varieties may be less sensitive to short-term fertilization differences and more strongly influenced by genetic traits that regulate reproductive development. While nutrient availability can influence yield components, the single-ear habit typical of many maize varieties may limit the extent to which external inputs can increase ear number under the tested conditions.

Table 2: Number of ears per plant in hybrid and local maize under different fertilizer treatments

Items	*Fertilizer Transactions*				*Mean*
	O_1	O_1*O_2	O_2	O_0	
Hybrid	1.66	1.66	1.33	1	1.41
	±0.47	±0.47	±0.47	0	
Local	1.33	1	1	1	1.08
	±0.47	0	0	0	
Mean	1.5	1.333	1.167	1	
L.S.D.$_1$	N.S				
L.S.D.$_2$	NS				
L.S.D.$_3$	NS				

O_1 = Organic fertilizer, O_2 =EM1, O_3 = Control, L.S.D. = Least Significant Difference at probability level0.05, L.S.D.$_1$ = Interaction, L.S.D.$_2$ = Fertilizer treatments, L.S.D.$_3$ = Cultivar, NS = Non-significant

Although ear number is an important yield component, several studies have shown that grain yield is not always directly correlated with the number of ears produced per plant (Sorro *et al*., 2015). Ear formation in maize is primarily determined by the genetic characteristics of the variety, with most cultivars producing only one to two ears per plant under normal conditions. This may explain the lack of significant differences observed among fertilizer treatments in the present study.

Several studies have demonstrated that the application of organic fertilizer significantly increased the number of ears per plant due to its role in improving soil properties and enhancing nutrient availability for plant uptake (Farhad *et al*., 2009). Similarly, Mohamed (2006) reported a significant increase in the number of ears per plant following the application of organic fertilizer, which may be attributed to the improvement in soil fertility and nutrient balance that supports better ear development. Although the current results showed only numerical increases rather than significant differences, the observed trend is consistent with earlier findings and suggests that organic and bio-enhanced fertilizers may contribute to improved ear formation under certain conditions.

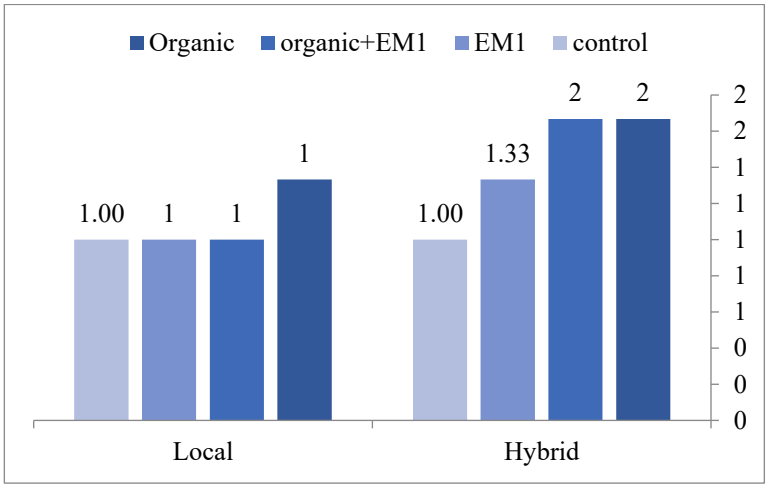

Figure 5: Effect of fertilizer treatments on the average number of ears per plant for hybrid and local maize cultivars

Figure 6: Effect of fertilizer treatments on the average number of ears per plant. (a) ears (hybrid variety), (b) mature ears (local variety), (c) Number of ears (local variety), (d) number of ears (hybrid variety)

Variance in Ear Diameter

Average Diameter of the Ear: The results presented in tables 3 and 4 and figure 7 show that fertilizer treatments had significant effects on the average ear diameter of both hybrid and local maize cultivars. For the hybrid cultivar, the EM1-organic fertilizer treatment recorded the highest average ear diameter (14.67 cm), followed by the organic treatment (14.50 cm), the EM1 treatment (12.50 cm), while the control recorded the lowest value (12.83 cm). Similarly, for the local cultivar, the EM1-organic treatment achieved the largest ear diameter (14.33 cm), followed by the EM1 treatment (13.33 cm), the organic fertilizer (12.67 cm), and the control (11.67 cm).

For minimum ear diameter, the hybrid cultivar showed no significant difference between the EM1 + organic and organic treatments, each recording the highest value (8.67 cm). These were followed by the EM1 treatment (7.67 cm), while the control recorded the lowest small ear diameter (6.33 cm). In the local cultivar, the EM1 + organic treatment produced the largest small ear diameter (8.33 cm), followed by the organic (7.67 cm)

and EM1 treatments (6.67 cm), whereas the control remained the lowest (4.67 cm). Overall, the results indicate that the use of EM1-organic fertilizers significantly enhanced ear diameter in both cultivars compared with the control, confirming their positive role in promoting maize growth and ear development.

Table 3: Maximum ear diameter (cm) of maize as affected by fertilizer treatments

Items	*Fertilizer Transactions*				*Mean*
	O_1	O_1*O_2	O_2	O_0	
Hybrid	14.5	14.67	12.5	12.83	13
	±1.41	±0.52	±1.02	±0.62	
Local	12.67	14.33	13.33	11.67	13.62
	±0.70	±0.40	±0.47	±0.94	
Mean	13.58	14.5	12.92	12.25	
L.S.D.$_1$	1.287				
L.S.D.$_2$	0.91				
L.S.D.$_3$	0.64				

Table 4: Minimum ear diameter (cm) of maize as affected by fertilizer treatments

Items	*Fertilizer Transactions*				*Mean*
	O_1	O_1*O_2	O_2	O_0	
Hybrid	8.67	8.67	7.67	6.33	7.83
	±0.65	±0.62	±0.47	±0.49	
Local	6.67	8.33	7.17	4.67	6.71
	±0.62	±0.49	±0.23	±0.528	
Mean	7.67	8.5	7.42	5.5	
L.S.D.$_1$	0.90				
L.S.D.2	0.641				
L.S.D.3	0.45				

O_1 = Organic fertilizer, O_2 =EM1, O_3 = Control, L.S.D. = Least Significant Difference at probability level0.05, L.S.D.$_1$ = Interaction, L.S.D.$_2$ = Fertilizer treatments, L.S.D.$_3$ = Cultivar

Variation in ear diameter is primarily attributed to differences in grain size and kernel fullness, while the cob diameter plays a crucial role in determining the total ear diameter (Khalifa and Al Hamad, 2008). Several studies have reported significant improvements in maize yield components, such as the number of ears per plant, ear length, and ear diameter, as a result of applying organic fertilizers (Mohamed, 2006).

The increase in ear diameter observed in the present study may be attributed to the application of organic fertilizers, particularly those containing chicken manure, which enhance nutrient availability and facilitate nutrient translocation within the plant (Anees, Khan and Ahmed, 2016). Organic fertilizers improve both the physical and chemical properties of the soil, thereby increasing nutrient uptake efficiency and promoting overall plant growth and development (Alamery and Alshatb, 2022). These findings are in agreement with those reported by (El-Sobky *et al.*, 2014) and (Omar, 2014), who also observed significant enhancements in ear diameter following the application of organic

fertilizers. The consistent results across studies further confirm the beneficial role of organic nutrient sources in improving maize ear development and yield performance.

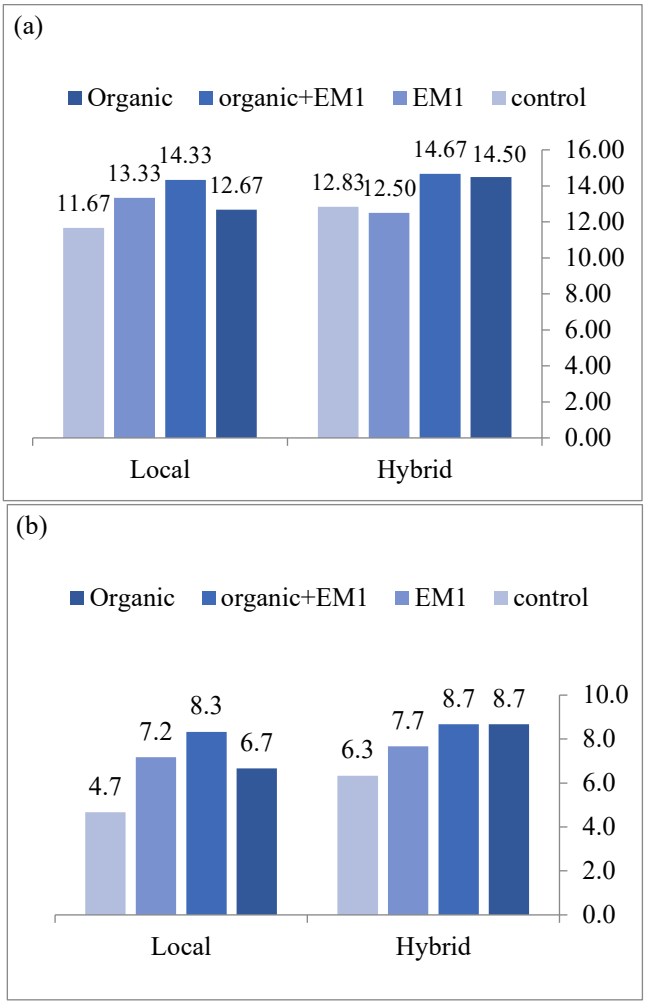

Figure 7: Effect of different fertilizer treatments on large (a) and small (b) ear diameter (cm) for hybrid and local maize cultivars

Number of Grains per Row

The results presented in Table 5 indicate significant differences in the average number of grains per row per ear for the two yellow corn cultivars, hybrid and local, under four fertilizer treatments: organic, bio-organic, biofertilizer, and control. For the hybrid cultivar, the bio-organic treatment recorded the highest average number of grains per row (53.33 grain row^{-1}), followed by the organic fertilizer (47.00 grain row^{-1}), biofertilizer (44.33 grain row^{-1}), while the control treatment gave the lowest value (39.67 grain row^{-1}). A similar trend was observed in the local cultivar, where the bio-organic treatment also produced the highest average (43.00 grain row^{-1}), followed by the organic fertilizer (37.67 grain row^{-1}), biofertilizer (35.00 grain row^{-1}), and the control treatment (30.33 grain row^{-1}). These results clearly demonstrate that the application of bio-organic fertilizers increased the number of grains per row in both cultivars compared to the

control, reflecting the positive effects of these fertilizers on ear development and kernel formation. The superior performance of the combined organic–EM1 treatment highlights the synergistic effect between organic matter and beneficial microorganisms in promoting reproductive growth and kernel formation.

Table 5: Number of grains per row in maize under different fertilizer treatments

Items	*Fertilizer Transactions*				*Mean*
	O_1	O_1*O_2	O_2	O_0	
Hybrid	47	53.33	44.33	39.67	46.08
	±4.24	±1.24	±0.471	±1.70	
Local	37.67	43	35	30.33	36.5
	±4.64	±2.16	±0.816	±4.64	
Mean	42.33	48.17	39.67	35	
L.S.D.$_1$	NS				
L.S.D.$_2$	4.62				
L.S.D.$_3$	3.27				

O_1 = Organic fertilizer, O_2 =EM1, O_3 = Control, L.S.D. = Least Significant Difference at probability level0.05, L.S.D.$_1$ = Interaction, L.S.D.$_2$ = Fertilizer treatments, L.S.D.$_3$ = Cultivar, NS = Non-significant

The increase in the number of grains per row, as shown in Figure 8, may be attributed to the role of organic matter in supplying essential nutrients such as nitrogen, phosphorus, and potassium, which enhance nutrient uptake and consequently improve grain set (Javaid and Mahmood, 2010). Moreover, the decomposition of organic matter results in the formation of organic acids capable of dissolving soil minerals and releasing available forms of potassium, thereby promoting better nutrient absorption and increasing the number of grains per row (Al-Zaidi, 2014).

The continuous availability of major nutrients (N, P, and K) from organic fertilizers throughout the plant's growth stages enhances leaf area expansion, dry matter accumulation, and overall plant vigor. These physiological improvements reduce ovary abortion and increase fertilization efficiency, leading to higher numbers of rows and grains per ear (Sharifi and Hizaden, 2009; Wuhaib, Al-Haidary and Makyia, 2009).

Similarly, the observed increase in grain number may be linked to the enhanced availability of nitrogen and other essential nutrients following the application of organic fertilizer, which supports vigorous plant growth and ear formation (Farhad *et al*., 2009). The combined use of organic fertilizers, poultry manure, and EM1 biofertilizers has been reported to increase the number of grains per row significantly (Fadlalla, Abukhlaif and Mohamed, 2016), while EM1 biofertilizers alone also exerted a positive effect on this trait (Tarang *et al*, 2013). These findings are consistent with the results reported by Farhad *et al*. (2009), Omar (2014), and El-Kholy *et al*. (2014), who demonstrated positive effects of organic and EM1 biofertilizer treatments on the number of grains per row. The improvement may also be attributed to the role of EM1 biofertilization in enhancing ear elongation by improving nutrient uptake, particularly nitrogen, which accelerates silk emergence and synchronizes it with pollen shedding, resulting in more efficient pollination and consequently a greater number of grains per row (Cirilo *et al*., 2009).

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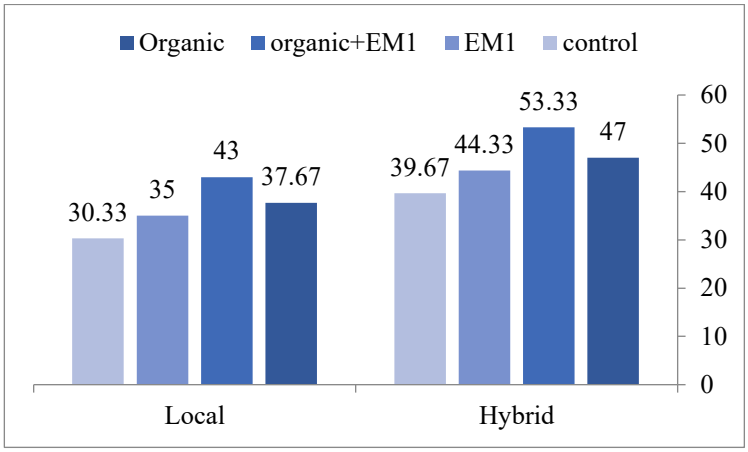

Figure 8: Average number of grains per single row in the ear for hybrid and local maize under different fertilizer treatments.

Effect of Fertilizer Treatments on Grain Quality

Grain protein content: The results presented in table 6 and figure 9 show that fertilizer treatments had a notable effect on grain protein content in both maize cultivars. For the hybrid cultivar, the bio-organic treatment recorded the highest average protein content (8.82 g), indicating its strong effectiveness in enhancing grain nutritional quality. This was closely followed by the organic fertilizer (8.81 g) and EM1 (8.79 g) treatments, while the control treatment produced the lowest protein content (8.24 g). These findings demonstrate that the hybrid variety consistently achieved higher protein content under all fertilized treatments compared with the control.

For the local cultivar, the EM1 + organic treatment resulted in the highest protein content (9.08 g), followed by the EM1 treatment (9.02 g). The organic fertilizer treatment recorded 8.77 g, whereas the control treatment produced the lowest value (8.38 g). Overall, the results indicate that both cultivars responded positively to organic and bio-organic fertilization, with the combined EM1-organic treatment showing the strongest enhancement in protein accumulation, particularly in the local variety.

Table 6: Grain protein content (g) of maize varieties under different fertilizer treatments

Items	*Fertilizer Transactions*				*Mean*
	O_1	O_1*O_2	O_2	O_0	
Hybrid	8.81	8.82	8.792	8.24	8.66
	±0.021	±0.011	±0.017	±0.001	
Local	8.77	9.08	9.026	8.38	8.81
	±0.013	±0.001	±0.001	±0.002	
Mean	8.79	8.95	8.90	8.31	
L.S.D.$_1$	0.010				
L.S.D.$_2$	0.007				
L.S.D.$_3$	0.005				

O_1 = Organic fertilizer, O_2 =EM1, O_3 = Control, L.S.D. = Least Significant Difference at probability level0.05, L.S.D.$_1$ = Interaction, L.S.D.$_2$ = Fertilizer treatments, L.S.D.$_3$ = Cultivar

The increase in protein content observed in the present study may be attributed to the role of decomposed organic matter in improving the soil's physical and chemical properties, thereby enhancing nutrient availability, particularly nitrogen, for plant absorption. Elevated nitrogen uptake directly increases nitrogen concentration in the grain, which in turn enhances protein synthesis (Al-Budeiri and Al-Shami, 2021). Several studies have consistently demonstrated that nitrogen is the primary determinant of protein content in maize grains (Triboi *et al*., 2000). These findings align with the present study, confirming that both organic fertilizers and EM1 biofertilizers enhance soil fertility and nutrient uptake, leading to improved protein content in maize grains.

Furthermore, incorporating organic matter into the soil can increase the availability of nitrogen, phosphorus, and potassium by increasing carbon dioxide, which forms H_2CO_3, in the soil solution (Rashwan, El-Sharkawy and Baddour, 2017). This is positively reflected in increased protein content. The application of EM1 also increased nitrogen accumulation in the grain by a percentage ranging from 1.31% to 3.43%, while in the year most favorable for corn production, the increase ranged from 3.58% to 18.19% (Cvijanović *et al.*, 2024).

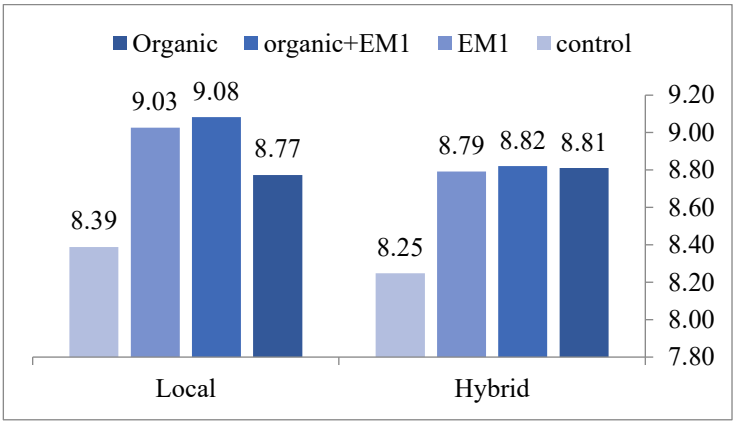

Figure 9: The effect of organic fertilizer and EM1 biofertilizer treatments on protein content (g) in two maize varieties (hybrid and local)

Grain Oil Content: The results presented in table 7 and figure 10 show that, in the hybrid variety, the combined organic + EM1 treatment produced the highest oil content at 8%, demonstrating its strong effectiveness in enhancing seed quality. This was followed by the organic treatment (7.5%), the EM1 treatment (7%), while the control recorded the lowest value (5.5%). For the local variety, the organic + EM1 treatment also resulted in the highest oil content (9%), indicating a similar positive response. This was followed by the organic treatment (6.50%), the EM1 treatment (6.50%), with the control again recording the lowest oil content (5.17%). Overall, the results clearly show that the integrated organic + EM1 treatment consistently enhanced oil accumulation in both the hybrid and local corn varieties compared to the control.

Table 7: Grain oil content (%) of maize varieties under different fertilizer treatments

Items	*Fertilizer Transactions*				*Mean*
	O_1	O_1*O_2	O_2	O_0	
Hybrid	8.50	8	7	5.50	7.25
	±0.40	±0.46	±0.12	±0.36	
Local	6	9	6.50	5.17	6.67
	±0.81	±0.08	±0.40	±0.12	
Mean	7.25	8.5	6.75	5.33	
L.S.D.$_1$	1.57				
L.S.D.$_2$	1.11				
L.S.D.$_3$	NS				

O_1 = Organic fertilizer, O_2 =EM1, O_3 = Control, L.S.D. = Least Significant Difference at probability level0.05, L.S.D.$_1$ = Interaction, L.S.D.$_2$ = Fertilizer treatments, L.S.D.$_3$ = Cultivar

These results can be explained by the fact that the use of organic fertilizer led to an increase in organic matter, availability of nutrients and nitrogen fixation by bioferment, as nitrogen is the basic substance in the formation of protein and microorganisms in the root zone that secrete plant hormones and substances that lead to increased growth and accumulation of dry matter, which in turn leads to an increase in oil concentration (El-Shafey and El-Hawary, 2016). Furthermore, EM1 biofertilization plays a key role in increasing leaf area and leaf area index, thereby improving the plant's efficiency in intercepting sunlight and enhancing photosynthetic activity. EM1 also enhances the plant's uptake of essential nutrients, including phosphorus (Heldt, 2005), which is required for the formation of ATP, the energy-rich compound essential for metabolic processes. Oil synthesis and storage require significantly more energy compared to protein formation; therefore, higher ATP production under EM1 treatment directly supports increased oil accumulation in corn grains. This explains the overall increase in oil percentage resulting from the positive effects of both organic fertilization and EM1 in improving grain yield and biochemical composition (Ibraheem, 2013).

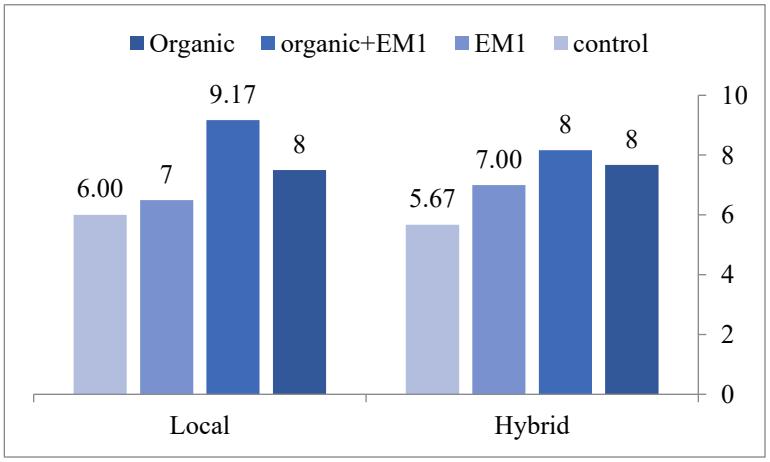

Figure 10: The effect of organic fertilizer and EM1 biofertilizer treatments on oil content (%) in two corn varieties (hybrid and local).

Conclusions

This study demonstrated that the combined use of organic fertilizers and EM1 significantly improved vegetative growth and grain quality in both hybrid and local maize varieties compared to the control treatment. The combined application of organic fertilizers and EM1 consistently yielded the best results. The combined application of organic fertilizer and EM1 increased the number of leaves, ear diameter, and number of grains per row, while also improving the oil and protein content of the grains. This demonstrates its effectiveness in enhancing both the yield potential and nutritional value of maize grains. Notably, the oil content in the grains reached its highest value in both varieties when organic fertilizer was applied with EM1, while the protein content in the grains increased significantly in the local variety, reflecting improved nutrient availability and uptake.

Based on these results, it can be concluded that combining organic fertilizers with EM1 represents a sustainable and effective strategy for nutrient management in maize production. This approach reduces reliance on chemical fertilizers, improves soil fertility and biological activity, and supports higher yields per unit area with a lower environmental impact. However, the study was conducted under specific soil and climatic conditions and during a single growing season. Although the biofertilizer (EM1) was used for the first time in the study area and showed clear and positive results compared to the control treatment, the study recommends further trials to monitor and evaluate the long-term effect of the biofertilizer under diverse environmental conditions.

Future research should focus on long-term field evaluations of organic fertilization and fertilization using the growth enhancer EM1, including their effects on soil health, crop stability, and economic yield. Further studies on different application rates and their integration with reduced chemical fertilizer use are also recommended to improve sustainable maize production systems.

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Authors' Declarations and Essential Ethical Compliances

Authors' Contributions (in accordance with ICMJE criteria for authorship)

Contribution	*Author 1*	*Author 2*	*Author 3*
Conceived and designed the research or analysis	Yes	Yes	Yes
Collected the data	Yes	Yes	Yes
Contributed to data analysis & interpretation	Yes	Yes	Yes
Wrote the article/paper	Yes	Yes	Yes
Critical revision of the article/paper	Yes	No	No
Editing of the article/paper	Yes	Yes	Yes
Supervision	Yes	No	No
Funding Acquisition	Yes	No	No
Overall Contribution Proportion (%)	40	30	30

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Research involving human bodies or organs or tissues (Helsinki Declaration)
The author(s) solemnly declare(s) that this research has not involved any human subject (body or organs) for experimentation. It was not clinical research. The contexts of human population/participation were only indirectly covered through literature review. Therefore, an Ethical Clearance (from a Committee or Authority) or ethical obligation of Helsinki Declaration does not apply in cases of this study or written work.

Research involving animals (ARRIVE Checklist)
The author(s) solemnly declare(s) that this research has not involved any animal subject (body or organs) for experimentation. The research was not based on laboratory experiment involving any kind animal. The contexts of animals were only indirectly covered through literature review. Therefore, an Ethical Clearance (from a Committee or Authority) or ethical obligation of ARRIVE does not apply in cases of this study or written work.

Research on Indigenous Peoples and/or Traditional Knowledge
The author(s) solemnly declare(s) that this research has not involved Indigenous Peoples as participants or respondents. The contexts of Indigenous Peoples or Indigenous Knowledge were only indirectly covered through literature review. Therefore, an Ethical Clearance (from a Committee or Authority) or prior informed consent (PIC) of the respondents or Self-Declaration in this regard does not apply in cases of this study or written work.

Research involving Plants
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